RESPONSE TO OFFICE ACTION APPL. No.: 10/533,611 DOCKET No.: TUV-031.01

## **AMENDMENTS**

## In the Specification:

Please delete the paragraph on page 2, line 31, to page 3, line 9, and replace it with the following paragraph:

Previously we have disclosed that with *B. mori* silk fibroin, a threefold helical polyglycine II or polyproline II type of conformation was stabilized by the interface, even though it is not observed in bulk. Valluzzi, R.; Gido, S. P. *Biopolymers* 1997, 42, 705-717; Valluzzi, R.; Gido, S.; Zhang, W.; Muller, W.; Kaplan, D. *Macromolecules* 1996, 29, 8606-8614; Zhang, W.; Gido, S. P.; Muller, W. S.; Fossey, S. A.; Kaplan, D. L. *Electron Microscopy Society of America, Proceedings* 1993, 1216. The *B. mori* fibroin crystallizable sequence is approximately (*Gly-Ala-Gly-Ser*)<sub>x</sub>, (SEQ ID NO: 4) and a left-handed threefold helical conformation, which is sterically reasonable, separates hydrophobic alanine and hydrophilic serine residues to opposite sides of the interface. Valluzzi, R.; Gido, S. P. *Biopolymers* 1997, 42, 705-717; Valluzzi, R.; Gido, S.; Zhang, W.; Muller, W.; Kaplan, D. *Macromolecules* 1996, 29, 8606-8614; Zhang, W.; Gido, S. P.; Muller, W. S.; Fossey, S. A.; Kaplan, D. L. *Electron Microscopy Society of America, Proceedings* 1993, 1216.

Please delete the paragraphs on page 7, line 11 to page 8, line 2, and replace them with the following paragraphs:

**Figure 9** depicts self-fabricated textured "tapes" from a peptide with sequence  $(Glu)_5(Ser-Gly-Ala-Gly-Val-Gly-Arg-Gly-Asp-Gly-Ser-GlyVal-Gly-Leu-Gly-Ser-Gly-Asn-Gly)_2(Glu)_5 (SEQ ID NO: 1). 1. Optical micrograph shows a ~10-15 micron texture which persists through the material thickness. The material is optically transparent. 2. Polarizing optical microscopy reveals patterned birefringence, indicating that the topographic texture is due to a changing material orientation. 3. SEM image shows the topographic structure of the tape. The difference in periodicity observed in SEM and optical microscopy is due to the fact that top$ 

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surface and bottom surface ridges are both observed in the optical image (resulting in an apparently shorter period).

Figure 10 depicts self-fabricated tapes of (Glu)<sub>5</sub>(Ser-Gly-Ala-Gly-Val-Gly-Arg-Gly-Asp-Gly-Ser-GlyVal-Gly-Leu-Gly-Ser-Gly-Asn-Gly)<sub>2</sub>(Glu)<sub>5</sub> (SEQ ID NO: 1) have "patterns within patterns" or a long-range ordered structure consisting of hierarchical nanoscale to microscale patterns; 1: the self-limited width and thickness of the fibers (~120 microns, 50 microns respectively) form the largest length scale in the hierarchy; a 40 micron periodic texture is observed running along the tape; 2: within the ridges of the 40 micron texture a 3 micron subtexture is observed; 3: a submicron texture of inclined sheets or layers can be observed (<40 nm, but exact size is below the resolution of the scanning electron microscope); TEM studies indicate a layer spacing of ~ 5nm.

Figure 11 depicts an IR spectra of self-fabricated tapes of (Glu)<sub>5</sub>(Ser-Gly-Ala-Gly-Val-Gly-Arg-Gly-Asp-Gly-Ser-GlyVal-Gly-Leu-Gly-Ser-Gly-Asn-Gly)<sub>2</sub>(Glu)<sub>5</sub> (SEQ ID NO: 1). Typically IR spectra for molecules are seen as very small differences in IR transmission relative to a large background, which must be subtracted out. Raw data (no background subtraction) is shown for transmission FTIR spectra through different regions (orientations) of the tape structure. Two orientations show very typical protein absorbance spectra over a high background. However in some orientations the IR radiation does not reach the detector.

Please delete the paragraph on page 9, line 13 and replace with the following paragraph:

Figure 25 depicts banded structures from engineered protein designed peptide (SEQ ID NO: 4).

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Please delete the paragraph on page 9, line 15 and replace with the following paragraph:

Figure 27 depicts spider silk modification. The images depict the air-water interface from an EDTA-Na aqueous solution of (Glu)<sub>5</sub>(Gly-Asp-Val-Gly-Gly-Ala-Gly-Ala-Thr-Gly-Gly-Ser)<sub>2</sub>(Glu)<sub>5</sub> (SEQ ID NO: 2).

Please delete the paragraph on page 9, line 18, and replace with the following paragraph:

Figure 30 depicts film morphology and helix anchoring: a) a designed helix with a stronger hydrophobic/hydrophilic difference will be more readily stabilized and anchored parallel to the interface; b) helical axis is perpendicular to smectic layer plane; and c) helices which tend to be parallel to interface and film result in layers more often normal to film.

Please delete the paragraph on page 28, lines 9-19 and replace with the following paragraph:

Desalted, HPLC purified, and lyophilized collagen-like peptide was obtained from the Protein Chemistry Core Facility at the Tufts Medical School. The sequence was (Glu)<sub>5</sub>(Gly-Val-Pro-Gly-Pro-Pro)<sub>6</sub>(Glu)<sub>5</sub> (SEQ ID NO: 3). The glutamic acid blocks were added to the ends of the peptides to promote solubility in water so that contaminant salts would not complicate analysis. Similar peptide design strategies have been used by Rotwarf et. al. to examine the solution behavior of  $\beta$ -sheet forming peptides. Rotwarf, D. M.; Davenport, V. G.; Shi, P.-T.; Peng, J.-L.; Sheraga, H. A. *Biopolymers* 1996, 39, 531-536. The collagen-like peptide was dissolved in 18 M $\Omega$  Millipore filtered water at a concentration of 1 mg/ml peptide in water. No salt or acid or extra reagent was required to aid dissolution. The solution was allowed to stand in an air-tight capped vial overnight, and then a gold mesh TEM grid (no substrate film) was dipped through the air-water interface.